Remarks

Claims 13, 14, 16-18, and 21-34 are canceled herewith without prejudice. Applicants reserve the right to file one or more continuing applications directed to the subject matter of the canceled claims.

New claims 35-51 are added. The new claims are submitted to more particularly point out and distinctly claim the subject matter which applicant believes is the invention. The new claims are submitted, in part, to avoid any confusion as to the subject matter being claimed in view of the prior prosecution by two different law firms. It is believed that the new claims more clearly point out the novel and non-obvious aspects of the claimed invention.

Support for the new claims is found at least in the original claims as filed and as discussed below.

Support for the limitations, "isolated" in the preamble and "wherein at least 96% of the bonds between the C and the B are in an L-configuration" in the body of the claim, is found at least on page 21, beginning on line 12 of the application which describes the purification procedure that was used to separate L-Pro-D-boroPro from L-Pro-L-boroPro:

> "High pressure liquid chromatography (HPLC) can be used to separate L-Pro-D-boroPro from L-Pro-L-boroPro. ... The L,L isomer comes off [the nucleosil C18 column] first at about 7 min., followed by the L,D isomer at about 10 min. NMR and mass spectra analysis were consistent with both compounds being Pro-boroPro. Rechromatography of the purified isomers indicated that the first pass on the HPLC column achieved *an isomeric purity of about 99-6% for each isomer*. High pressure liquid chromatography (HPLC) can similarly be used to separate L-Ala-D-boroPro from L-Ala-L-boroPro *or to separate the D-boroPro form of other inhibitors ([of DPIV] from the L-boroPro form.*"

Support for the limitation, "wherein the compound inhibits DPIV activity", is found in the preamble of original claim 13, i.e., "[a]n inhibitor of DPIV".

Support for the limitation, "wherein X comprises an amino acid or a peptide", also is found

10. X is peptide, provided that the compound inhibits DPIV activity (the original preamble limitation).

Support for the limitation of new claim 48, wherein X is the {A-proline-A`-}$_m$ moiety also is found in now-canceled claim 13.

No new matter has been added.

New claim 35 is a Generic Claim and embraces the previously elected Species

Applicants previously elected the specific compound L-Pro-L-boroPro as the species for search purposes only and stated that "upon allowance of a generic claim which embraces this species, Applicants reserve their right to consideration of claims to additional species which are written in dependent form or otherwise include all of the limitations of an allowed generic claim" (response filed Feb. 20, 1998).

New claim 35 is a generic claim which embraces the elected species. Accordingly, Applicants respectfully request examination of the generic claim with respect to the elected species and that the Examiner consider all of the newly submitted claims upon allowance of this generic claim.

Rejection of Claim 23 Under 35 U.S.C. §112(1)

Claim 23 is rejected under 35 U.S.C. §112(1) for recitation of the phrase, "substantially pure".

Claim 23 is canceled herewith.

The new claims recite limitations that are based upon the specific language that appears on page 21 (line 12) of the specification, namely, that the isomeric purity of the compound is from about 96-99 percent.

In view of the foregoing, applicants respectfully request that the Examiner find that the new claims satisfy the requirements of 35 U.S.C. §112(1).

Objection to the Specification and Rejection of Claims Under 35 U.S.C. §112(1)

The specification is objected to under 35 U.S.C. §112(1) for failing to provide an enabling disclosure. Claims 13, 14, 16, and 21-28 are rejected under §112(1) for the reasons set forth in the Objection to the Specification. The Examiner takes the position that the effects of varying the N-terminus amino acid are unknown and that substitution of peptides bearing an Asp, Glu, Arg, or Lys residue at the N-terminus are "likely to be inactive". The Examiner further states that if the second amino acid from the C-terminus is important to enzyme recognition, "loss of activity is likely with these embodiments".

Claims 13, 14, 16, and 21-28 are canceled herewith.

Two groups of new claims are added. The "Group A" claims (claims 35-41) are directed to an isolated compound that is a *dipeptide* ("wherein A′ comprises an amino acid") containing the boroPro moiety. The "Group B" claims (claims 42-51) are directed to an isolated compound that is a *dipeptide* ("wherein X comprises an amino acid") or a polypeptide ("wherein X comprises ... a peptide"). Each of the Group A and Group B claims includes the further functional limitation that the isolated compound "inhibits DP-IV activity".

The specification provides working examples which describe the synthesis and isolation of two exemplary compounds which fall within the scope of the new claims. Further compounds which differ from those specifically described in the specification differ only in the nature of the amino acid or peptide that is covalently coupled via a peptide bond to the boroProline moiety. Accordingly, *making* additional compounds which are defined by the structures shown in the new claims involves substituting one or more amino acids for those shown in the working examples and following the peptide synthesis procedures described in the specification. Peptide synthesis is routine in nature. Accordingly, *making* the compounds embraced by new claims 35-41 (dipeptides) and new claims 42-51 (peptides containing two or more amino acids) does not require "undue" experimentation.

The specification also describes assays to determine whether a compound "inhibits DP-IV

"undue" experimentation because compounds which "inhibit DP-IV activity" can be selected by using any of a variety of functional assays which measure DP-IV activity and determining whether the putative inhibitor inhibits this activity. According to the specification (e.g., see description beginning on page 16, line 11), various types of assay systems can be used to determine whether the claimed compounds act inhibit DP-IV activity. For example, a particularly simple and straight-forward test for measuring a DP-IV inhibitory activity that is an enzyme inhibitory activity is provided on page 19 (line 1) of the specification. This paragraph describes a spectrophotometric assay in which DP-IV enzyme activity is measured as release of a chromatographic product. As stated in the description of this assay, "[r]eduction in activity in the presence of inhibitor provides an easy test for inhibitory activity". In this manner, compounds which fall within the scope of new claims 35-51 can be tested for DP-IV inhibitory activity. Such screening assays are routine in nature and do not amount to undue experimentation.

In view of the foregoing, it is believed that the rejection of claim under 35 U.S.C. 112(1) based upon undue experimentation is not applicable to the new claims. Accordingly, applicants respectfully request that the Examiner find that the new claims satisfy the enablement requirement of 35 U.S.C. 112(1).

Rejection of Claims 13, 14, 16, 21 and 22 Under 35 U.S.C. §102(e)

Claims 13, 14, 16, 21 and 22 are rejected under 35 U.S.C. §102(e) as being anticipated by Bachovchin, U.S. 4,935,493 (Bachovchin '493).

Claims 13, 14, 16, 21 and 22 are canceled herewith.

The isolated compounds of new claims 31-51 are not taught in Bachovchin '493. Accordingly, Applicants respectfully request that the Examiner find the new claims patentable over Bachovchin '493.

Rejection of Claims 13, 14 and 16 Under 35 U.S.C. §102(a)

Bachovchin, J. Biol. Chem. 265:3738 (1990) ("Bachovchin JBC 1990").

Claims 13, 14 and 16 are canceled herewith.

The isolated compounds of new claims 31-51 are not taught in Bachovchin JBC 1990. Accordingly, Applicants respectfully request that the Examiner find the new claims patentable over Bachovchin '493.

Rejection of Claims 23 - 25 under 35 U.S.C. §103

Claims 23-25 are rejected under 35 U.S.C. §103 as being unpatentable over the Bachovchin reference (J. Biol. Chem. 265, 3738, 1990). The Examiner directs Applicant's attention to Bachovchin Table I, page 3740 as depicting "several compounds falling within the scope of claim 23." The Examiner states that this reference does not employ the phrase "substantially pure preparation" but contends that "the organic chemist of ordinary skill is acutely aware that upon synthesizing a given compound, some impurities are nearly always present prior to further purification" and that the "presence of impurities, together with the compound of interest, gives rise to a 'substantially pure preparation.'"

Claims 23-25 are canceled herewith. New claims 35-51 are added. Each of the new claims includes the limitation that "at least 96% of the bonds between the C and the B are in an L-configuration".

The Bachovchin (JBC 1990) reference does not teach or suggest a composition as now claimed in the new claims, namely, wherein "at least 96% of the bonds between the C and the B are in an L-configuration". Although the reference states that attempts were made to further purify the various isomers present in the reaction mixture (p. 3743), such purification attempts were *unsuccessful*. At the time the Bachovchin (JBC 1990) reference was submitted for publication, the authors *incorrectly presumed* that one of the peaks collected following chromatography on silica gel represented an enriched fraction of the L-boro Pro isomer (see p. 3743). Dr. Bachovchin subsequently discovered that the peak which had been *presumed* to be

discussed in the following paragraphs.

The *only* description of any type of purification of the stereoisomers in the JBC reference is found in the paragraph which describes the H-Ala-boroPro-pinacol preparation (page 3743):

> "NMR analysis indicates that this column [silica gel] partially separates the two isomers of ala-boroPro-pinacol. The early fraction appears from the NMR spectra to be approximately 95% enriched in one isomer. Because this early fraction has more inhibitory power than the later fractions at equal concentrations, we *presume* that early fraction is enriched in the L-boroPro isomer ... further characterization of the isomers based on stereo specific synthesis will be published in a separate paper."

Although the JBC reference "*presumed*" that the early fraction eluted from the silica gel column is Ala-boroPro-pinacol, there is no basis in the cited art to support this presumption. At the time the JBC reference was submitted, the authors, including Dr. Bachovchin, did not fully appreciate the multiple forms (e.g., geometric isomers, optical isomers, intramolecular reaction products) in which the dipeptides could exist (see, e.g., W. Gutheil and W. Bachovchin, "Separation of L-Pro-DL-boroPro into Its Component Diastereomers and Kinetic Analysis of Their Inhibition of Dipeptidyl Peptidase IV. A New Method for the Analysis of Slow, Tight-Binding Inhibition", Biochemistry 32: (1993) (copy enclosed). This later publication describes the preparation of L-L and L-D Pro-boroPro Diastereomers by C18 HPLC as described in the legend of Fig. 2. These conditions are substantially identical to the HPLC separation conditions provided in the pending application.

The Biochemistry reference further illustrates the structure of trans-Pro-boroPro and shows its chiral centers (figure 1). According to this reference, the coupling of L-Pro with racemic LD-boroPro is expected to yield a mixture of two diastereomers: L-Pro-L-boroPro and L-Pro-D-boroPro. Figure 1 does not illustrate the potential cis form of these stereoisomers. According to this reference, the absolute configurations of the purified isomers were designated on the basis of a

Bachovchin's JBC reference was submitted, the authors had not performed a sufficiently detailed NMR analysis to determine, in fact, whether the two peaks eluted from the silica gel column represented, for example, the two optical isomers (diastereomers) of Ala-boroPro or whether the two peaks represented the cis and trans forms of one stereoisomer or a mixture of the cis and trans forms of the two different optical isomers. Absent the detailed NMR analysis, one could not definitively conclude that the "early fraction" of the silica gel column described in the JBC reference represented the L isomer.

Moreover, at the time Bachovchin JBC (1990) was published, the authors did not appreciate that Ala-boroPro could undergo an intra molecular reaction to form a cyclic (a less active) intermediate. This undesired side reaction is described in the pending application:

> "The two diastereomers of Ala-boroPro-pinacol, L-Ala-D-boroPro-pinacol and L-Ala-L-boroPro-pinacol, can be partially separated by silica gel chromatography with 20% methanol in ethyl acetate as eluant. The early fraction appears by NMR analysis to be 95% enriched in one isomer. Because this fraction has more inhibits [sic] DP-IV to a greater extent than later fractions (at equal concentrations) it is probably enriched in the L-boroPro (L-Ala-L-boroPro-pinacol) isomer."

Bachovchin JBC (1990) describes the preparation of H-Ala-boroPro-pinacol from the 50/50 diastereomeric mixture on page 3743. Preparation of the Ala-boroPro-pinacol was accomplished by deblocking the amino terminal at 0°C with a 3.5 molar excess of HCL-dioxane and allowing the reaction to continue at 0°C for 15 minutes, followed by *room temperature incubation for one hour*. Subsequent purification was attempted on a silica gel column with 20% methanol in ethyl acetate as eluant. Under these conditions, it is believed that H-Ala-boroPro may have undergone an intra molecular reaction to form a six-membered ring and, potentially, additional side reactions. Accordingly, the "early" and "later" fractions described in the Bachovchin (JBC 1990) reference could have represented the linear and cyclic forms, respectively, of the H-Ala-boroPro dipeptide. This would explain the reduced activity of the "later" fraction compared to that of the "early"

In summary, the authors of the Bachovchin (JBC 1990) reference ultimately *abandoned silica gel chromatography* as an approach for purifying the stereoisomers and, instead, developed an alternative reverse phase technique (the C18 separation described in the pending application and in Dr. Bachovchin's 1993 Biochemistry reference) to separate the purified L isomers. Dr. Bachovchin never concluded precisely why the silica gel separation technique had been unsuccessful; however, in view of the foregoing, one can reasonably conclude that the "early" and "later" fractions which had been eluted from the silica gel column had not, in fact, represented a separation of the optical isomers. *Upon request by the Examiner, Applicants can submit additional NMR results in the form of a declaration by Dr. Bachovchin to evidence that the optically pure L-isomer is different from the compound that reportedly was presumed to be the L-isomer in Bachovchin JBC (1990).* These results would further evidence that the Bachovchin (JBC 1990) reference does not teach or suggest the claimed L isomers.

In view of the foregoing, Applicants respectfully request that the Examiner reconsider and find new claims 35- 51 patentable over Bachovchin (JBC 1990).

Rejection of Claims 18, 23-28 and 30 Under 35 U.S.C. §103

Claims 18, 23-28 and 30 are rejected under 35 U.S.C. §103 as being unpatentable over Bachovchin (USP 4,935,493). The Examiner asserts that this reference does not employ the phrase "substantially pure preparation" but that the organic chemist of ordinary skill would recognize that some impurities are nearly always present prior to further purification and that the presence of such impurities gives rise to a "substantially pure preparation." The Examiner also asserts that the attainment of a substantially pure preparation would have been obvious to the pharmaceutical chemist of ordinary skill.

Claims 18, 23-28 and 30 are canceled herewith. New claims 35- 51 are added. Each of the newly added claims includes the limitation that "at least 96% of the bonds between the C and the B are in an L-configuration".

isomers that may be present in the preparation. Thus, the Bachovchin '493 patent does not render obvious the claimed isolated isomers. Moreover, in view of the facts presented in the preceding paragraphs regarding the rejection of claims under the Bachovchin (JBC 1990) reference, namely, the difficulties presented in isolating the claimed isomers, there is no basis in the Bachovchin '493 patent to overcome this deficiency in the reference. Thus, even if one of skill in the art had been motivated to further purify the various isomers of the claimed compounds (which Applicants dispute), there is no knowledge in the art cited by the Examiner to suggest to one of ordinary skill in the art how this objective could be achieved. Further, there is no knowledge in the art to suggest that any one particular isomer was more active than another isomer. It was not until Applicants discovered the enhanced activity and correlated the enhanced activity to the particular isomer form claimed in this application that one would have been: motivated to develop methods to purify said compounds in view of the (inaccurate) report of the isomer purification in Bachovchin (JBC 1990). As discussed above, the authors of the Bachovchin (JBC 1990) reference *had incorrectly assumed* that a particular peak fraction which eluded from a silica gel chromatography column was enhanced in the L-isomer. Accordingly, the Bachovchin (JBC 1990) reference does not teach, suggest, or render obvious the invention as now claimed.

In view of the foregoing, Applicants respectfully request reconsideration and find new claim 35- 51 patentable over the Bachovchin '493 patent.

Rejection of Claims 13, 14, 16, 18, 21-28 and 30 under 35 U.S.C. §103

Claims 13, 14, 16, 18, 21-28 and 30 are rejected under 35 U.S.C. §103 as being unpatentable over Bachovchin (WO 89/03223) or Flentke (PNAS 88, 1556, 1991) or Metternich (USP 5,288,707). The Examiner states that each of the Bachovchin WO 89/03223, Flentke and Metternich publications "teach compounds falling within the scope of the claimed genus." No further explanation is provided to support the assertion that the claims are "rendered obvious."

Claims 13, 14, 16, 18, and 21-23 are canceled herewith. New claims 35- 51 are added.

the C and the B are in an L-configuration".

The arguments presented above in regard to the Bachovchin JBC 1990 reference and Bachovchin '493 patent are reiterated here. None of the additional references cited by the Examiner for this rejection of claims under 35 U.S.C. §103 cures the deficiencies in the teachings of the primary reference. None of these references, alone or in combination, teach or suggest a composition that contains a compound having the formula recited in the new claims, wherein at least 96% of the bonds between the C and the B are in an L-configuration.

Bachovchin WO 89/03223 does not teach or suggest the purification of an L-isomer. Rather, this Bachovchin PCT publication suggests separating the isomers using a silica gel filtration column, the *same* column described in Bachovchin (JBC 1990) that *failed to separate* the isomers. Accordingly, the Bachovchin WO 89/03223 publication does not teach, suggest or render obvious the invention as now claimed.

The Flentke reference refers acknowledges the Bachovchin (JBC 1990) reference for the procedures for preparing the compounds discussed in Flentke. The arguments presented above are reiterated here, namely, this reference does not teach, suggest or render obvious the invention of new claims 35-51. There is no further suggestion in the Flentke publication of a method for isolating the claimed compound in which "at least 96% of the bonds between the C and the B are in an L-configuration".

The Metternich patent describes boro lysine compounds which are "potent thrombin inhibitors." The boro lysine compounds are structurally distinct from and have different functional activities compared to the claimed boroProline compounds. Accordingly, this patent is irrelevant to the issue of obviousness of the claimed composition. Applicants suspect that the Examiner may have cited this reference because Metternich refers to the existence of an asymmetric carbon atom as the carbon which is attached to the boron moiety (column 4, line 35). However, the Metternich teachings with respect to the existence of such an asymmetric carbon atom are limited to stating merely that the carbon atom "may have the D- or L-configuration, or

isomer is preferred over the other. Accordingly, there is no motivation provided by Metternich to one of ordinary skill in the art to purify any particular isomer and, further, there is no teaching or suggestion in Metternich of the manner in which the L and D isomers could be separated from one another. In view of the foregoing, there is no basis upon which to find that Metternich renders obvious the claimed compounds which differ structurally and functionally from the Metternich boroLysine compounds.

In view of the foregoing, Applicants respectfully request that the Examiner find new claims 35 - 51 patentable over the prior art.

Information Disclosure Statement Comments

The Examiner indicated certain deficiencies in the Information Disclosure Statement, e.g., incomplete citations and ambiguity as to whether the complete or a part of the reference had been provided. In view of the Examiner's comments, a revised Information Disclosure Statement is being prepared to make clear the articles that were considered by the Examiner.

Summary

Applicants wish to expedite the prosecution of this application. Accordingly, if the Examiner feels that a telephone conference would be helpful, he is respectfully requested to call the undersigned attorney at the telephone number presented below.

It is believed that the rejections of record are not applicable to the pending claims. Accordingly, it is respectfully requested that favorable action on the new claims be taken.

Respectfully submitted,

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